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First record of plastic ingestion by a freshwater stingray

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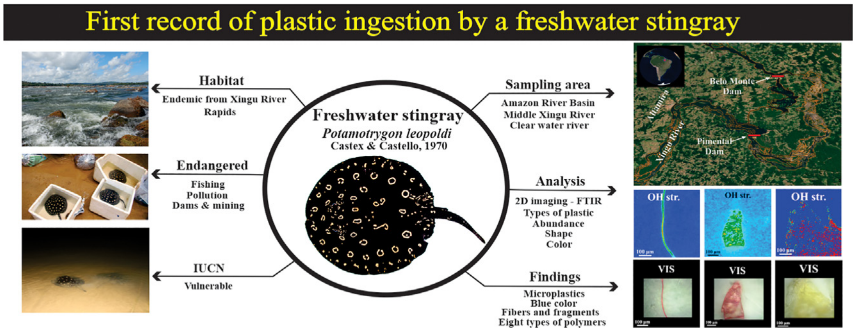
HIGHLIGHTS

- First report of plastic ingestion by freshwater elasmobranchs in the world
- 66.6 % of gastrointestinal tracts sampled contained plastic particles.
- Particle type was dominated by fibers (64.2 %) and blue color (33.3 %).
- The most frequent polymer was artificial cellulose fiber (59.7 %).

GRAPHICAL ABSTRACT



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ABSTRACT

The abundance and dispersion of plastic particles in aquatic ecosystems has become pervasive resulting in the incorporation of these materials into food webs. Here we describe the first record of plastic ingestion by the freshwater white-blotched river stingray *Potamotrygon leopoldi* (Potamotrygonidae), an endemic and threatened species in the Xingu River, Amazon basin. Potamotrygonidae stingrays inhabit exclusively Neotropical rivers, occupying rocky substrate habitats and feeding mainly on benthic macroinvertebrates. The gastrointestinal tract of 24 stingrays were analyzed, 16 (66.6 %) of which contained plastic particles. In total, 81 plastic particles were recorded and consisted of microplastics (< 5 mm, n = 57) and mesoplastics (5–25 mm, n = 24). The plastic particles found were classified into fibers (64.2 %, n = 52) and fragments (35.8 %, n = 29). The predominant color was blue (33.3 %, n = 27), followed by yellow (18.5 %, n = 15), white (14.8 %, n = 12), black (13.6 %, n = 11), green (6.2 %, n = 5), transparent (4.9 %, n = 4), pink, grey and brown (2.5 %, n = 2, each) and orange (1.2 %, n = 1). No significant correlation was observed between the number of plastic particles and the body size. Eight types of polymers were identified in the plastic particles analyzed using 2D FTIR Imaging. The most frequent polymer was artificial cellulose fiber. This is the first report of plastic ingestion by freshwater elasmobranchs in the world. Plastic waste has become an emerging problem in aquatic ecosystems globally and our results provide an important datapoint for freshwater stingrays in the Neotropics.

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1. Introduction

Environmental pollution by synthetic polymer particles — which include microplastics (< 5 mm), mesoplastics (5 to 25 mm) and macroplastics (≥ 25 mm) (Barrows et al., 2018) — is an emerging problem, which has the potential to disrupt aquatic ecosystems around the world (Gall and Thompson, 2015; Worm et al., 2017; Blettler and Wantzen, 2019). Due to their longevity, widespread dispersal, negative impacts, and pervasiveness, plastic particles are a threat to freshwater and marine biota (Gall and Thompson, 2015; Alimba and Faggio, 2019; Argeswara et al., 2021).

It is important to understand the effects on aquatic biota from various types of plastics, in a variety of structures (i.e., fibers and particles) and sizes (i.e., micro to macro) (Blettler and Wantzen, 2019). Plastic entanglement and ingestion can cause acute harm of the digestive and respiratory tracts, as well as chronic physiological and toxicological damage (e.g., Azevedo-Santos et al., 2021). Assessments of plastic ingestion in freshwater ecosystems have so far been limited when compared to studies carried out in the marine environment (Blettler and Wantzen, 2019), as a result, direct comparisons, for example, between plastic ingestion by various taxa are impossible (Santos et al., 2021).

Most studies on plastic ingestion have focused on marine teleost fish (e.g., Bray et al., 2019; Neto et al., 2020; López-Martínez et al., 2021; Savoca et al., 2022; Fabri-Ruiz et al., 2023). Regarding elasmobranchs, previous work has been focused on sharks as well as pelagic or benthic marine rays (Germanov et al., 2019; Parton et al., 2020; Pegado et al., 2021; Lipej et al., 2022; Pinho et al., 2022; Gong et al., 2023). To date, there is no data on the plastic ingestion by freshwater elasmobranchs around the world (see Azevedo-Santos et al., 2019, 2021; Monteiro et al., 2022).

Plastic pollution in freshwater ecosystems has been the subject of studies globally (Cera et al., 2020; Bellasi et al., 2020; Azevedo-Santos et al., 2021). Recently, there has been increased attention within the Amazon basin concerning the increase in plastic pollution in the natural environment (Giarrizzo et al., 2019). Ingestion of synthetic polymers has been reported for >200 freshwater species, most of these are fish (Azevedo-Santos et al., 2021). For example, 50 species have been identified from Brazilian inland waters to ingest plastics (Lima et al., 2021) — a notable exception to this list are elasmobranchs.

The white-blotched river stingray *Potamotrygon leopoldi* Castex & Castello, 1970, is a freshwater species endemic to the Xingu River basin. This species is long-lived, has a slow growth rate, late sexual maturation, low fecundity and long gestation periods, thus presenting a reduced ability to recover from both acute and chronic negative impacts throughout ontogeny. It is considered vulnerable to anthropic impacts due to its restricted distribution and increasing degradation of its habitats (Charvet et al., 2005). It is a species with great international appeal within the ornamental fish trade, as it is among the most commercialized Potamotrygonidae species in Brazil, being exported to several countries in the world, mainly to the Asian, European, and North American (Charvet et al., 2018). It is listed as "Vulnerable" by the International Union for the Conservation of Natural Resources – IUCN (IUCN, 2023), and this status is justified due to the use of limited natural distribution, increasing pollution, loss of habitat associated with illegal mining and to the construction of the Belo Monte Hydroelectric Plant on the Xingu River (Sanches et al., 2021).

Considering the increasing amount of plastic released into the environment, especially in freshwater ecosystems (Slootmaekers et al., 2019; Cardozo et al., 2023), we expected that Neotropical stingrays are increasingly exposed to plastic pollution and, therefore, possibly this contamination causes both physiological and toxicological impacts on these sensible species soon. In this context, to improve conservation policies for this species and its habitats, it is important to assess whether white-blotched river stingrays are exposed to plastic pollution and based on their feeding behavior if this species can serve as an indicator of plastic pollution in the environment. This study aims to investigate the occurrence of plastic particles in the gastrointestinal tract of *P. leopoldi*; characterize plastic particles in terms of abundance, diversity, shape, color and composition of synthetic polymers ingested by the species and correlate the size and weight of specimens with plastic contamination. This is the first evidence of plastic ingestion by a freshwater stingray in the world.

2. Material and methods

2.1. Study area

The Xingu River is approximately 2300 km long and originates in the state of Mato Grosso and enters the Amazon River at river kilometer (RKM) 420 (Camargo and Júnior, 2008). Due to its underlying geology, it is classified as a "clearwater" tributary (Lucas et al., 2021). The climate is typical of equatorial rainforest and considered tropical, hot and humid, with an average annual temperature ranging from 17.5 to 24.5 °C, and average annual precipitation ranging from 2.066 mm to 2.379 mm (Camargo et al., 2015). The section of the river studied is known as the middle Xingu River, it is approximately 300 km long and includes the lower Iriri River at its confluence with the Xingu River (03°49'16.7" S and 052°38'32.7" W), the Bacajá River (03°42'04.2" S and 051°32'54.1" W), and downstream to the Belo Monte dam (02°07'50.9" S and 051°39' 44.4" W) (Fig. 1). Within this reach, the river is characterized by heterogenous aquatic habitats including bedrock, sand, reservoir, and floodplain with dynamic seasonal hydrology determined by wet and dry seasonality (Zuluaga-Gómez et al., 2016).

2.2. Sampling procedures and samples processing

Specimens of *P. leopoldi* were captured by longlines and cast nets between April 2020 and October 2021. All individuals were fixed in 10 % formalin and transported to the laboratory. The disc length (DL), disc width (DW) and total weight (W) of each specimen were measured. Gastrointestinal tracts were collected via a longitudinal incision in the abdominal region, using surgical forceps and a scalpel. The gastrointestinal tracts were carefully removed, and their contents placed in Petri dishes for analysis under a stereomicroscope (Nikon SMZ800N) at 8 × to 32 × magnification. All the plastic particles identified during this analysis were separated and carefully placed in Petri dishes containing distilled water. Particles were photographed, measured, and sorted according to shape and color (Hidalgo-Ruz et al., 2012). The estimates of plastic ingestion quantified in our study may be underestimated. In the analytical method adopted our lower size limit of particles recovered was 0.8 mm, which was established on the smallest size we were comfortable handling with tweezers.

2.3. Control of contamination of plastic particles in the environment

We adopted and eventually adapted procedures recommended as described and reviewed by Prata et al. (2021) in order to provide reliable results about microplastics contamination in experiments. Our analysts used cotton lab coats and gloves straight from packaging during all experiments. The only people in the room were analysts, keeping doors and windows closed and air conditioning turned off. Materials and equipment used during the laboratory processing were cleaned before and after each procedures analysis with distilled water and protected with aluminum foil or glass lid. We use alcohol (70 %) filtered through a 1 μm porosity mesh to store the gastrointestinal tract and analyze their content. Each plastic particle found was placed in an Eppendorf (sterilized) with distilled water for later analysis. The procedures were done under a hood with air extractor that was cleaned daily and Petri dishes that had the gastrointestinal tracts were covered with a glass lid when they were not moved and only open if necessary. Three clean Petri dishes with distilled water were placed next to the stereomicroscope during each analysis of gastrointestinal tracts and inspected after each sample processing to identify possible external contamination. Even after all minimization procedures, microplastics of the environment may be found in the samples. To account for this potential contamination, at the end of each gastrointestinal tracts analysis, all Petri dishes were analyzed in the hood and, in the case of plastic particles observed, those were compared to the plastic particles found in the samples,

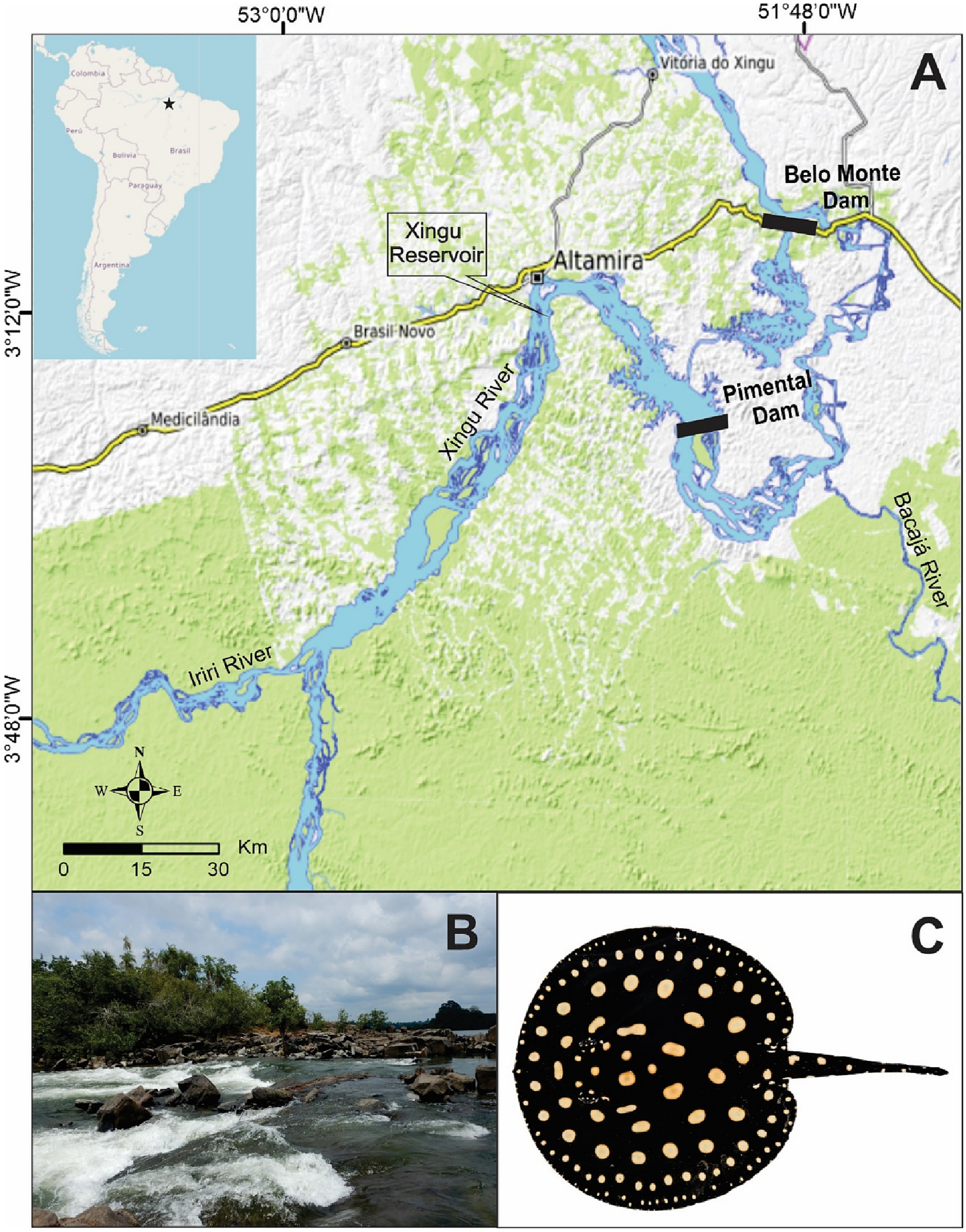

Fig. 1. Sampling area of the freshwater stingray *Potamotrygon leopoldi* in the Middle Xingu River, Pará, Brazil (A). Lotic environment in the Middle Xingu River, habitat of the species *P. leopoldi* (B). Image of the freshwater stingray *P. leopoldi* (C). Credit to Dr. Leandro Melo for the photo by *P. leopoldi*.

if they were of the same type, color and similar size, plastic particles of the sample were discarded.

2.4. Analysis

The percentages of the different categories of shape and color, as well as the polymeric composition of the particles, and the frequency of occurrence (FO%) of the plastics found in the gastrointestinal tracts were enumerated. The FO% was calculated by: $FO\% = (Ni / N) \times 100$, where Ni = the number of gastrointestinal tracts that contained plastic particles, and N = total number of gastrointestinal tracts examined.

Kendall's rank correlation was performed to assess the association between the size (disc width - DW) or weight (W) of stingray's specimens and the number of plastics found, to determine whether the plastic ingestion changes throughout ontogeny.

Samples of each category of plastic found in the rays' gastrointestinal tracts were separated for 2D imaging - Fourier transform infrared (FTIR) analysis (Su et al., 2016). FTIR analysis was conducted directly on, dry filters (without further processing) using a Cary 620–670 FTIR microscope equipped with a 128 × 128 FPA detector (Agilent Technologies). Procedural blanks for plastic contamination (e.g., airborne plastics) were carried out to check for laboratory contamination and interferences. The spectra were recorded directly on the surface of the samples (or on the gold background) in reflectance mode, with an aperture and spectral resolution of 8 cm^{-1}, with 128 scans being acquired for each spectrum. A "single tile" analysis resulted in a 700 × 700 μm^2 (128 × 128 pixels) map, with each image map having a spatial resolution of 5.5 μm (i.e., each pixel has an area of 5.5 × 5.5 μm^2) (Pegado et al., 2021). The small pixel size allowed for the collection of numerous independent spectra on each plastic sample, e.g., >150 independent spectra are typically collected on a 1 mm long and 10 μm thick fiber (Morais et al., 2020). Diagnostic bands (at least 2–5) of the fibers/fragments were identified and matched with those of references reported in the literature, taking care to check the full spectral profile of each polymer (Jung et al., 2018). The series of plastic samples found in the gastrointestinal tracts (Figs. 3 and 4), consisted of fragments (identified as blends of cellulose, polyurethane, acrylate and polyamide, see below) that were large enough (hundreds of microns) to allow their feasible analysis through Attenuated Total Reflectance (ATR), using an ATR slide with a germanium crystal mounted on the FTIR microscope (Garside and Wyeth, 2003).

3. Results and discussion

A total of 24 specimens of *P. leopoldi* were collected and analyzed. Size (disc length) of the examined *P. leopoldi* individuals ranged from 34.4 cm to 71.2 cm (mean 50.0 cm ± 12.0 SD) and the mean disc width ranged from 33.5 cm to 64.5 cm (mean 46.9 cm ± 9.7 SD; Table 1). Sixteen of the 24 gastrointestinal tracts analyzed (66.6 %) had plastic particles from which a total of 81 plastic particles were recovered. Specimens that contained plastic particles averaged 3.8 (SD ± 1.6) particles per individual and

Table 1
Biometrics (Disc Length – DL, Disc Width – DW, and Total Weight – TW) of freshwater stingray *Potamotrygon leopoldi* specimens and characteristics (Type, Shape, Color, Polymer type) of the plastic particles found in their gastrointestinal tract. The presence of plastic particles (PoP) was expressed as presence (1) or absence (0), as well as the Number (N) of plastic particles recovered from each gastrointestinal tract. The types are Mi = Microplastic; Me = Mesoplastic. The polymers are AC = Artificial Cellulose; PS = Polystyrene; ABS = Acrylonitrile Butadiene Styrene; PA = Polyamide; Teflon; PET = Polyethylene Terephthalate; PU = Polyurethane; Acrylate.

Stingray	DL (cm)	DW (cm)	TW (g)	PoP	Type	Shape	Color	N	Polymer
1	34.4	33.5	1700	1	Mi	Fiber	Black	1	AC
					Mi	Fiber	Blue	1	AC
					Me	Fiber	Black	1	AC
					Me	Fiber	Orange	1	AC
2	41.7	39.4	2900	0	–	–	–	0	–
3	43.4	40	3400	0	–	–	–	0	–
4	71.2	64.5	14,000	0	–	–	–	0	–
5	49.5	48.7	5600	1	Mi	Fiber	Blue	2	–
6	35	34	1800	1	Mi	Fiber	Blue	3	AC
7	34.5	35.5	1500	1	Mi	Fiber	Blue	1	AC
8	69	55	13,666	1	Mi	Fragment	White	2	PS
					Mi	Fiber	White	2	AC
					Mi	Fragment	Brown	1	PA
9	35	36	2450	1	Mi	Fiber	Black	3	Teflon
					Mi	Fiber	Yellow	4	AC
10	47	48	5800	1	Mi	Fragment	Green	1	–
11	56.9	49.4	8900	1	Mi	Fiber	Blue	3	AC, Teflon
					Mi	Fiber	Pink	1	AC
					Me	Fiber	Blue	1	AC, Teflon
					Me	Fragment	Grey	2	AC, Blend (PA, PU)
					Me	Fragment	Yellow	4	PA, Blend (PA, PU)
					Me	Fragment	Transparent	1	Blend (PA, PU)
					Me	Fragment	Blue	1	ABS
12	37.2	55	3200	1	Me	Fiber	Blue	1	AC
					Mi	Fiber	Blue	3	AC
13	38.9	36.5	2100	1	Mi	Fiber	Blue	1	AC
					Mi	Fiber	Green	1	AC
14	58.5	52	8100	0	–	–	–	0	–
15	56.5	50.7	6300	1	Mi	Fiber	Black	2	AC
					Mi	Fiber	White	1	ABS
					Mi	Fiber	Blue	1	AC
					Me	Fragmento	White	2	ABS
16	66.9	60.2	12,500	1	Mi	Fiber	Blue	1	AC
17	68.9	64.4	12,500	0	–	–	–	0	–
18	69.4	63.9	13,500	1	Mi	Fiber	Black	3	–
					Mi	Fiber	Green	3	–
19	48.8	42.5	4000	0	–	–	–	0	–
20	50	46.8	5000	0	–	–	–	0	–
21	41	35.4	2950	1	Mi	Fragment	Transparent	1	–
					Mi	Fragment	White	3	–
					Mi	Fragment	Blue	1	–
22	45.8	41.4	3900	1	Mi	Fiber	Black	1	PET
					Mi	Fiber	Transparent	1	AC
					Mi	Fiber	Blue	1	AC
					Mi	Fiber	White	1	PET
					Me	Fragment	Brown	1	PA
					Me	Fragment	Transparent	1	PA
					Me	Fragment	White	1	PET
					Me	Fragment	Yellow	6	Blend (PA, PU)
23	54.3	49.6	6200	0	–	–	–	0	–
24	47.4	44.7	4100	1	Mi	Fiber	Blue	4	AC
					Me	Fiber	Blue	1	AC
					Mi	Fiber	Blue	1	AC
					Mi	Fiber	Yellow	1	AC
					Mi	Fragment	Pink	1	AC, Blend (ABS + Acrylate)

ranged from one to twelve particles. Most plastic particles were classified as microplastics (70.4 %; n = 57), followed by mesoplastics (29.6 %; n = 24). The mean size of the microplastics found was 3.1 mm (SD ± 1.2) and ranged from 0.8 to 4.7 mm. Whereas the mesoplastics mean size was 8.2 (SD ± 3.6) mm, ranging from 5.2 to 13.7 mm. Plastic types consisted of 64.2 % (n = 52) fiber, and 35.8 % (n = 29) fragments. The predominant color was blue (33.3 %; n = 27), followed by yellow (18.5 %; n = 15), white (14.8 %; n = 12), black (13.6 %; n = 11), green (6.2 %; n = 5), transparent (4.9 %; n = 4), pink, grey and brown (2.5 %; n = 2, each) and orange (1.2 %; n = 1) (Figs. 2 and 3). Although non-significant, positive correlations were detected between the number of plastic particles and the size (p = 0.674) and weight (p = 0.726) of the specimens. These findings suggest that there is no bioaccumulation of plastic particles in gastrointestinal tract of the rays as smaller, presumably younger, rays have statistically similar numbers of microplastics as larger, older rays.

The freshwater stingray *P. leopoldi* is a benthic predator that feeds on a variety of prey items including fish and mollusks (Shibuya, 2022). This makes this group of fish susceptible to the bioaccumulation of

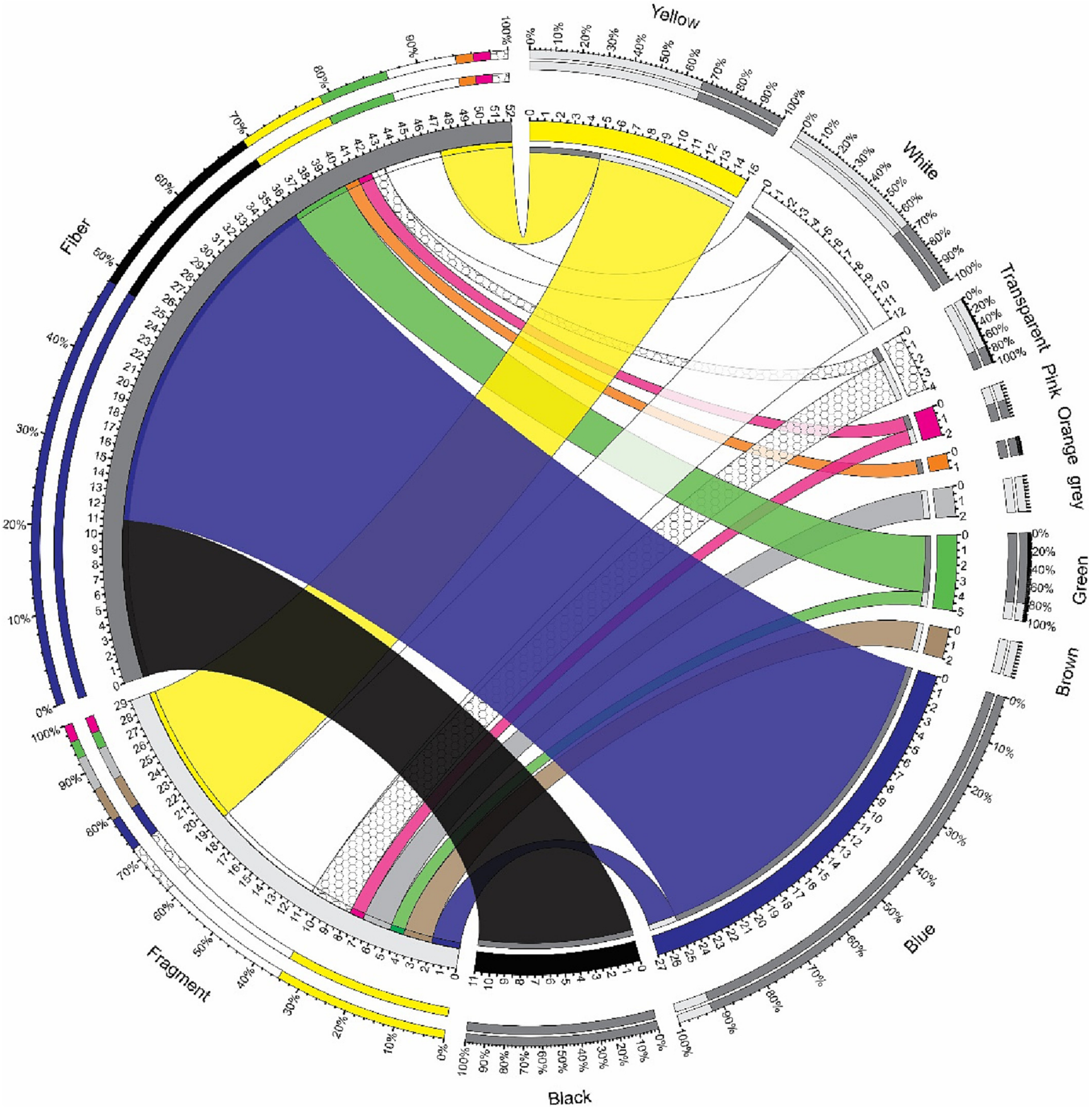

Fig. 2. Chord diagram showing the classification of plastics found in the gastrointestinal tract of freshwater stingray *Potamotrygon leopoldi* collected in the Middle Xingu, Pará, Brazil. The colors shown in the figure represent the colors of the recovered plastic particles. For each color, the number and percentage between fiber and fragment was calculated. The dark grey color represents the fiber, and the light grey color represents the fragment. The percentage values represent the proportion between each format (fiber or fragment) and the color of the plastic particles. And the numbers represent the amount of plastic particles found for each shape and color. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

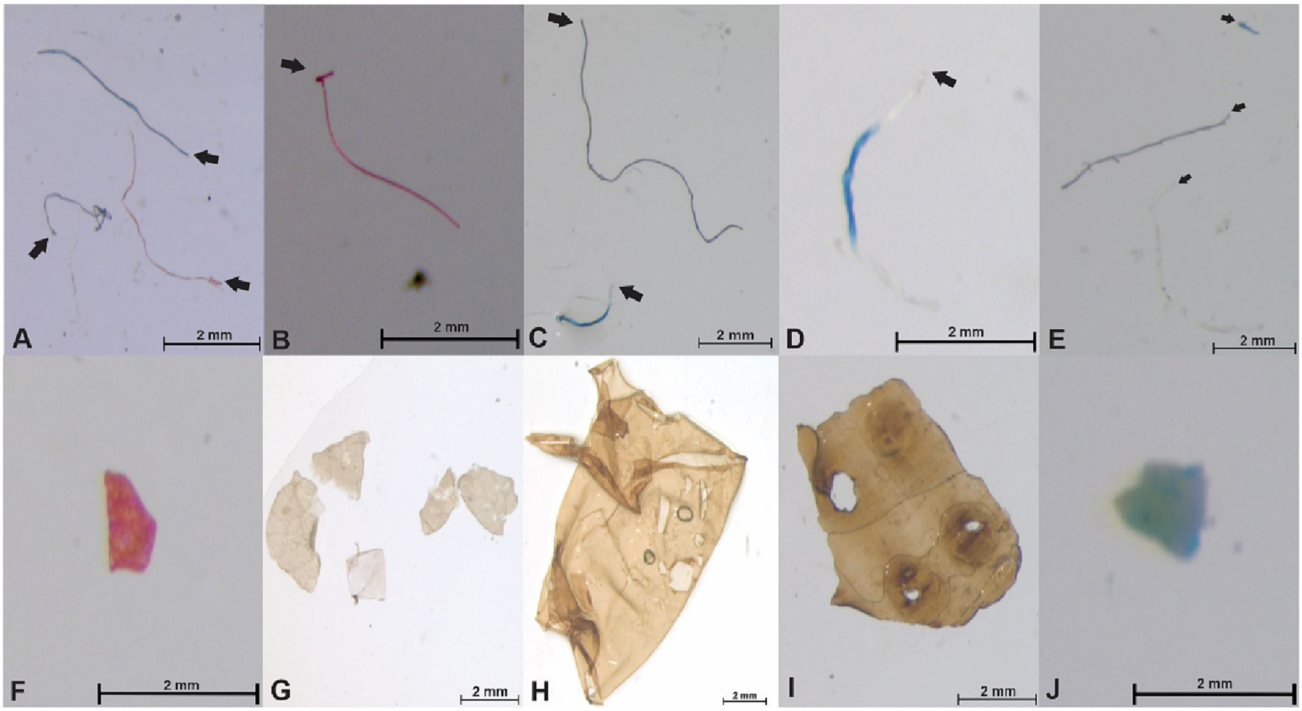

Fig. 3. Plastics found in the stomach contents of freshwater stingray *Potamotrygon leopoldi* specimens collected in the Middle Xingu River, Pará, Brazil. From A to E are the fibers and the different colors. From F to J are the fragments and colors. The black arrows indicate the location of the fibers.

contaminants affiliated with plastic particles that are incorporated into lower trophic levels. Therefore, the high prevalence of plastic particles, which may be associated with the foraging strategy of this group of fishes (Jabeen et al., 2017; Pinho et al., 2022), highlights the potential vulnerability of elasmobranchs to plastic pollution (Consales and Marsili, 2021).

Plastic particles, once ingested, can be expelled, or accumulated in the gastrointestinal tract of elasmobranchs (Fossi et al., 2018; Germanov et al., 2019; Valente et al., 2019). Among the main physical impacts of ingestion non-natural food resources are blockage of the digestive tract, translocation to other tissues and mechanical damage (Derraik, 2002; Jovanović, 2017). In addition, other negative health effects are associated such as decreased growth rate, reduced fertility, energy depletion, increased immune response, feeling of satiety leading to starvation, decreased food consumption and weight loss (Sussarellu et al., 2016; Herrera et al., 2019; Consales and Marsili, 2021). Investigations in marine environments have shown that persistent organic pollutants contained in water and sediment can be adsorbed by microplastics and, therefore, can become pollutant transfer vectors, generating endocrine disruption as well as additional health effects (Rochman et al., 2014; Wardrop et al., 2016). It is possible that freshwater stingray species are exposed to the same risks as marine species, mainly because they are predatory species, they are susceptible to greater risk. Therefore, further studies are needed to assess the impacts of plastic pollution on this iconic group of fish.

The high prevalence of fibers found in gastrointestinal tracts were similar to other studies of marine stingrays (De Lucia et al., 2014, 2018; Rochman et al., 2014; Neves et al., 2015; Alomar et al., 2016; Gianeti et al., 2019; Consales and Marsili, 2021; Argeswara et al., 2021; Pegado et al., 2021). The distribution of plastic particles in rivers is influenced directly by anthropogenic processes. From the largest cities to the smallest indigenous villages, most Amazon settlements are in riparian areas along >80,000 km of navigable waters (Giarrizzo et al., 2019). The region's torrential rains, coupled with increasingly frequent and severe floods, and lack of wastewater treatment can wash plastic waste into streams and rivers (Giarrizzo et al., 2019).

The plastic waste is trapped within flooded forests, therefore degrading into microplastics that can easier be incorporated into the soil and/or carried back into the water (Gonçalves et al., 2020). Studies in the Middle Xingu River and the Amazon estuary revealed microplastic particles in the digestive tracts of 13 freshwater and 14 marine fish species, 20 of which are commonly consumed by humans (Pegado et al., 2018; Andrade et al., 2019). Once ingested by a fish, these microplastics may then be transported to muscle or other tissues, where the particles may be retained for the fish's entire lifespan (Karami, 2017).

The dominance of blue plastics is interesting and worth expanding — at least locally — a discussion already raised by other authors (Urbanski et al., 2020; Okamoto et al., 2022; Coelho et al., 2023). Previous work showed a prevalence of black particles in the fish assemblage of Xingu River (Andrade et al., 2019). This contrasts with the predominance of blue particles described in this study. It is possible that *P. leopoldi* or their prey preferentially ingest blue plastic due to potential attraction to that color, but more study is needed to confirm this.

Eight types of polymers were identified in the plastic particles analyzed by 2D FTIR Imaging (Fig. 4; Table 1). The most frequent polymer was artificial cellulose fibers (59.7 %), followed by blend of Polyamide (PA) and Polyurethane (PU) (13.4 %), Teflon (10.4 %), Polyamide (PA; 9 %), Acrylonitrile Butadiene Styrene (ABS; 6%b), Polyethylene Terephthalate (PET; 4.5 %), Polystyrene (PS; 3 %) and blend of ABS and Acrylate (1.5 %). Cellulose was found only in the procedural blanks (Garcia et al., 2021). Artificial cellulose (e.g., Rayon and other industrial textiles) has recently come to attention as a pollutant along with other synthetic fibers (Savoca et al., 2019; Macieira et al., 2021), as it can transfer dyes and additives that are harmful to aquatic fauna. Therefore, its potential impact on protected species should be the topic of future studies. The second most common polymers were a blend of PA and PU, which could come from fishing gear, like nets and floats that are often have these polymers in their composition and contain a petrochemical-derived polymer (Collard et al., 2018). ABS has a wide range of application in electronic industries, light industries, automobile industries, etc., due to its excellent mechanical properties such as toughness and good impact strength, good chemical resistance, ease of molding, processing advantages and high-quality surface finishing (Ma et al., 2012; Xu et al., 2012). PET is one of the polymers most produced by industries, worldwide (Andrady, 2011). This polymer is used in the

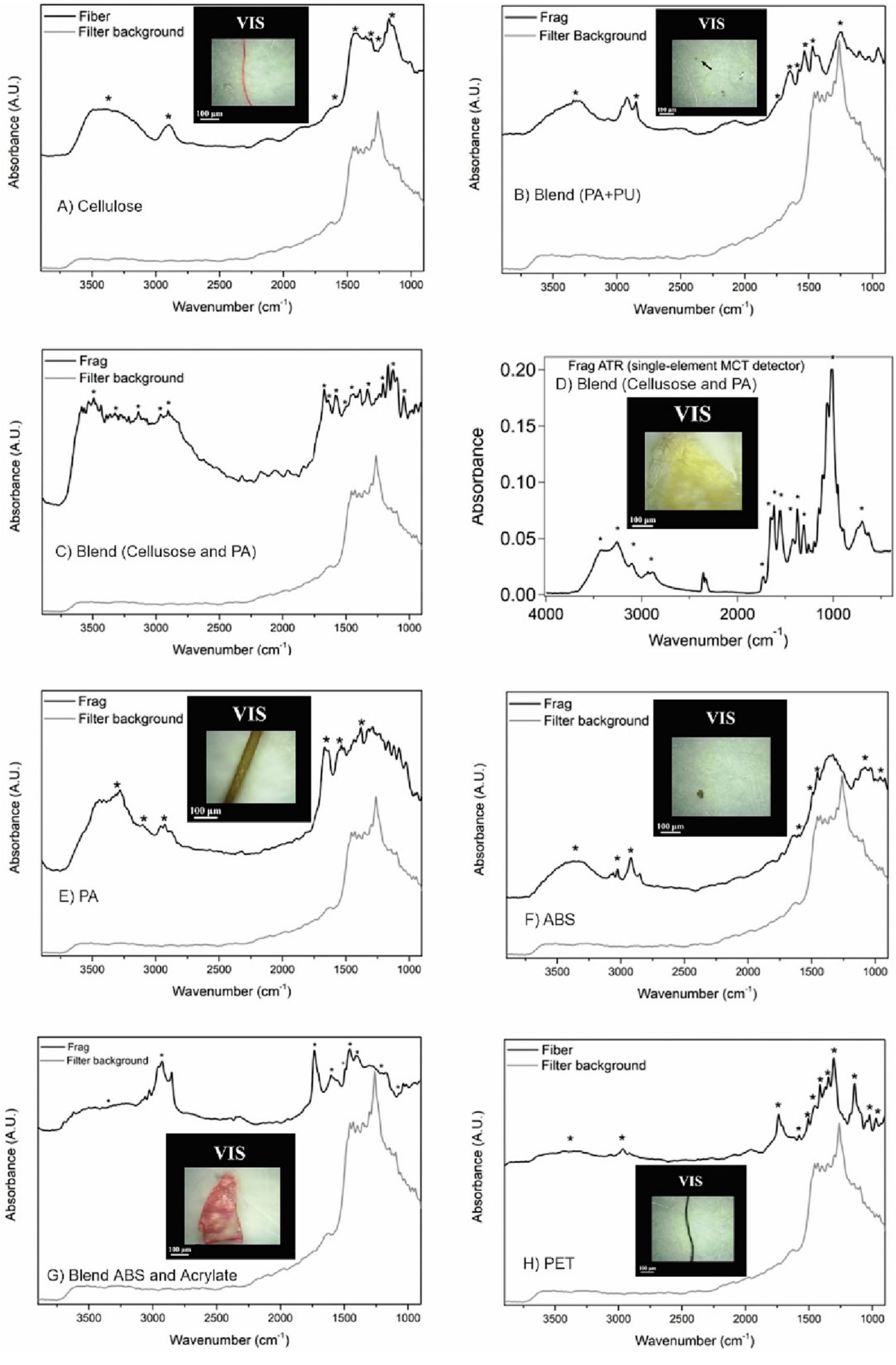

Fig. 4. Representative FTIR reflectance spectra acquired different plastics polymers, collected from the stomach contents of the freshwater stingray *Potamotrygon leopoldi* from Xingu River, Pará, Brazil. A) Artificial Cellulose; B) Blend (PA + PU): Polyamide + Polyurethane; C) Blend Cellulose and PA; D) Blend Cellulose and PA with fragment ATR; E) PA: Polyamide; F) ABS: Acrylonitrile Butadiene Styrene; G) Blend of ABS and Acrylate; and H) PET: Polyethylene Terephthalate.

production of textiles, including clothes, blankets, and fleeces, as well as bottles (Wang et al., 2014). PET fibers are common in domestic wastewater, often from clothes washing machines, which are ultimately discharged into rivers and eventually oceans (Browne et al., 2011; Napper and Thompson, 2016). As a relatively dense polymer, PET is also more likely to sink to the bottom of aquatic environments, where it can be ingested by benthic organisms (Ghosal et al., 2018), including the *P. leopoldi*. PS is a colorless transparent polymer used in household applications, electronics, packaging, insulation foam, single use items like disposable cutlery, etc. (Kaloyianni et al., 2021). Due to the lightness of most of its products, it is very difficult to recycle and often ends up in the environment, contributing significantly to the formation of microplastics (MPs).

4. Conclusions

This study is an important step towards a better understanding of contamination status of freshwater stingrays in the neotropical region and the risks it represents. Plastic particles were found in the gastrointestinal tract of two-thirds of *P. leopoldi* specimens analyzed. The size and weight of stingrays did not influence the number of ingested particles. This shows how much this species, at all stages of life, is exposed to plastic pollution. *P. leopoldi* is an iconic and important target for ornamental fishing in the Amazon River basin and is among the most exported freshwater stingrays in Brazil. Most of the particles found were fibers, and the most frequent polymer was the artificial pulp fibers. Our study provides the first record of plastic ingestion by a freshwater stingray worldwide, as well as an important baseline for future comparisons of exposure of this group of elasmobranchs to plastic contaminants in the freshwater environment. The lack of knowledge about the impacts of plastics on these organisms reflects the need for more research to be conducted to better understand the effects of plastic pollution on neotropical stingrays. Investigations, especially for areas and species still little studied, are important contributions to understanding spatial and temporal patterns of plastic pollution in ecosystems and aquatic organisms, as well as to encourage effective prevention and conservation efforts in response to this global problem.

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CRediT authorship contribution statement

Paulo A.A. Trindade: Conceptualization, Methodology, Formal analysis, Visualization, Writing – original draft, Writing – review & editing. **Lúcio D.M. Brabo:** Investigation, Data curation, Visualization, Writing – review & editing. **Ryan Andrades:** Investigation, Data curation, Visualization, Writing – review & editing. **Valter M. Azevedo-Santos:** Writing – review & editing. **Marcelo C. Andrade:** Writing – review & editing. **Laura Candore:** Investigation, Data curation, Writing – review & editing. **Serena B. Cabigliera:** Investigation, Data curation, Writing – review & editing. **David Chelazzi:** Investigation, Data curation, Writing – review & editing. **Alessandra Cincinelli:** Investigation, Data curation, Writing – review & editing. **Carson A. Jeffres:** Supervision, Writing – review & editing. **Tommaso Giarrizzo:** Conceptualization, Funding acquisition, Project administration, Supervision, Visualization, Formal analysis, Methodology, Writing – review & editing.

Data availability

The authors do not have permission to share data.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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